Supplementary Information

N/A- Provided as separate Excel files

Figure S1.

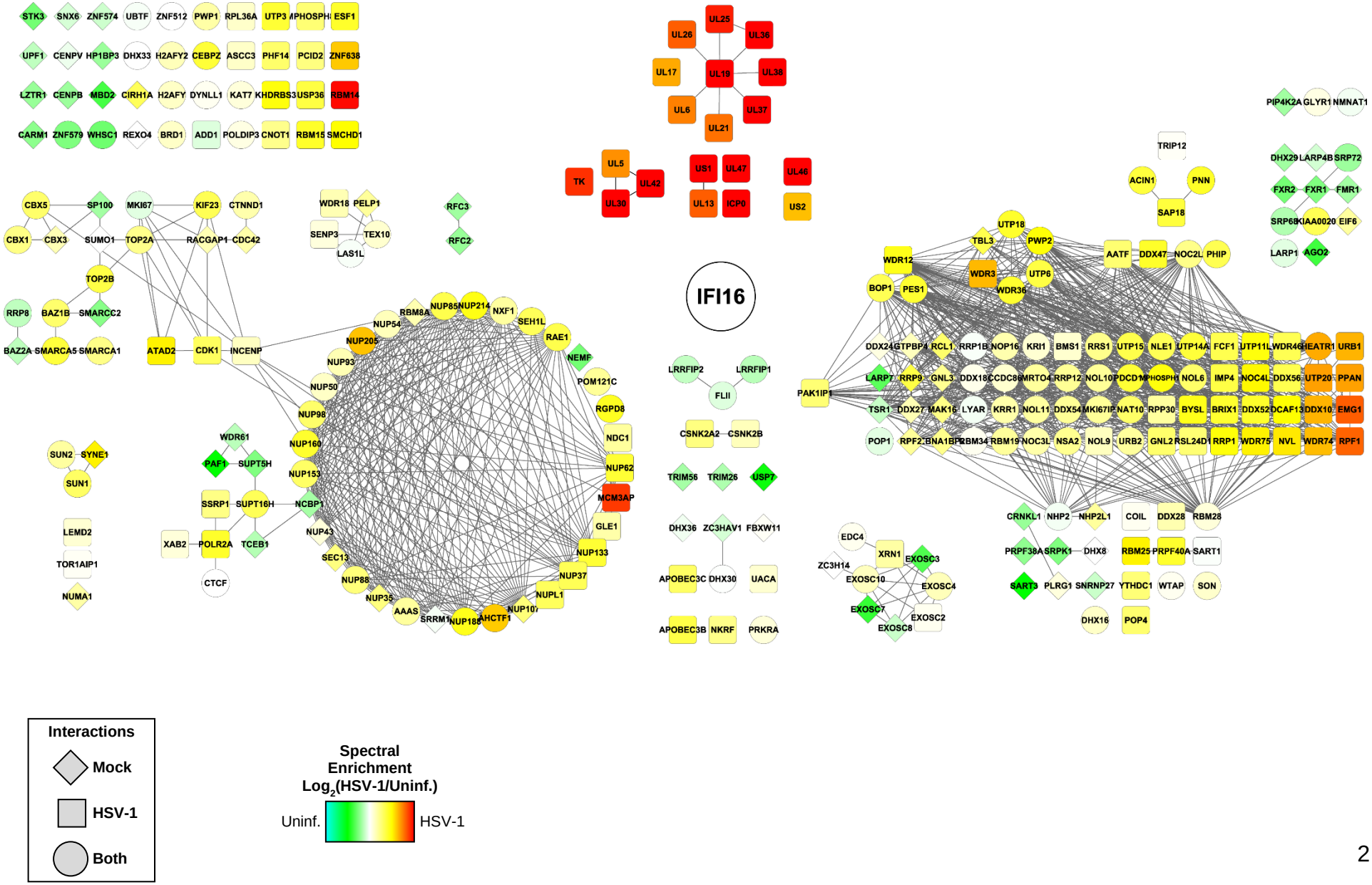
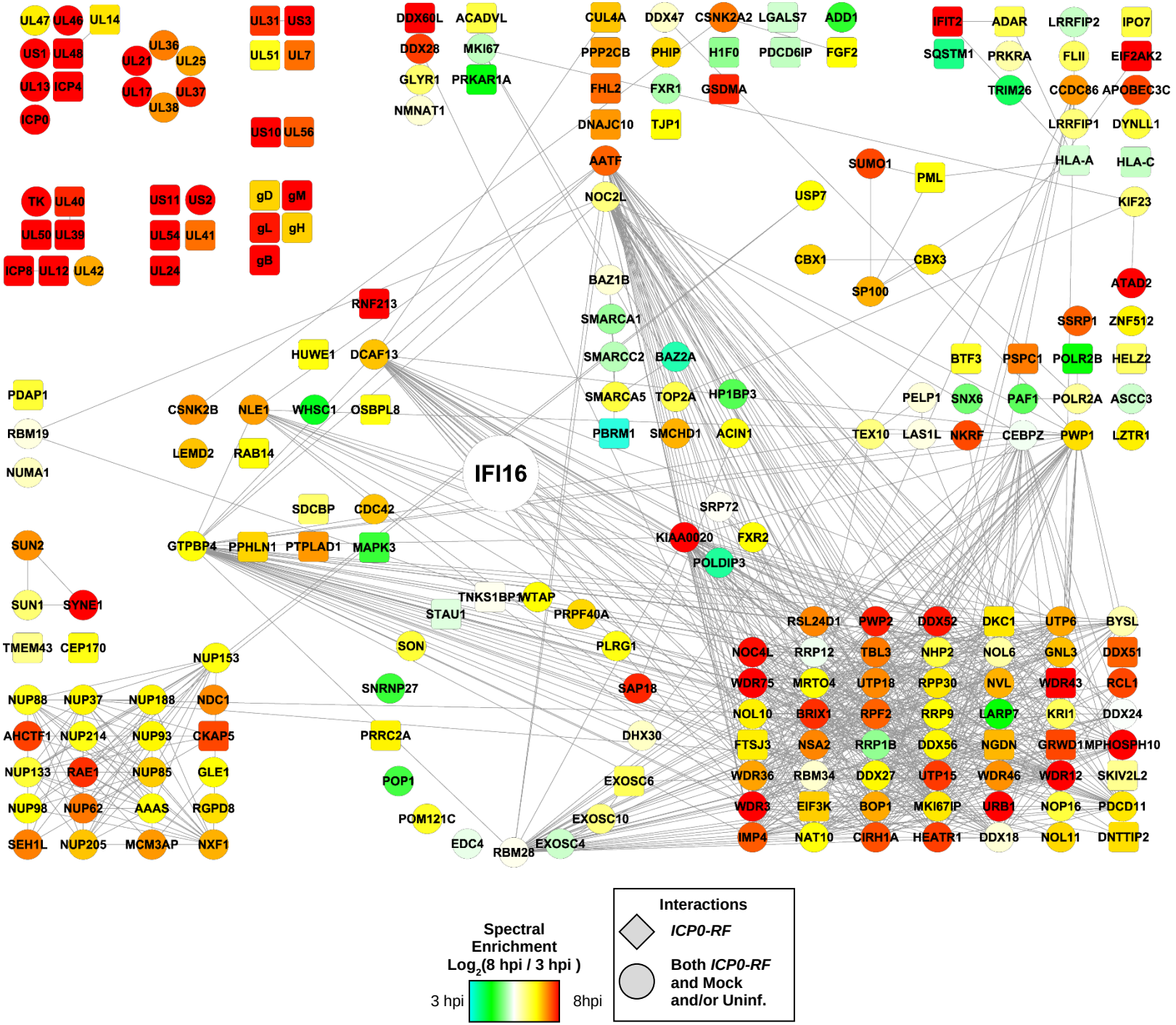
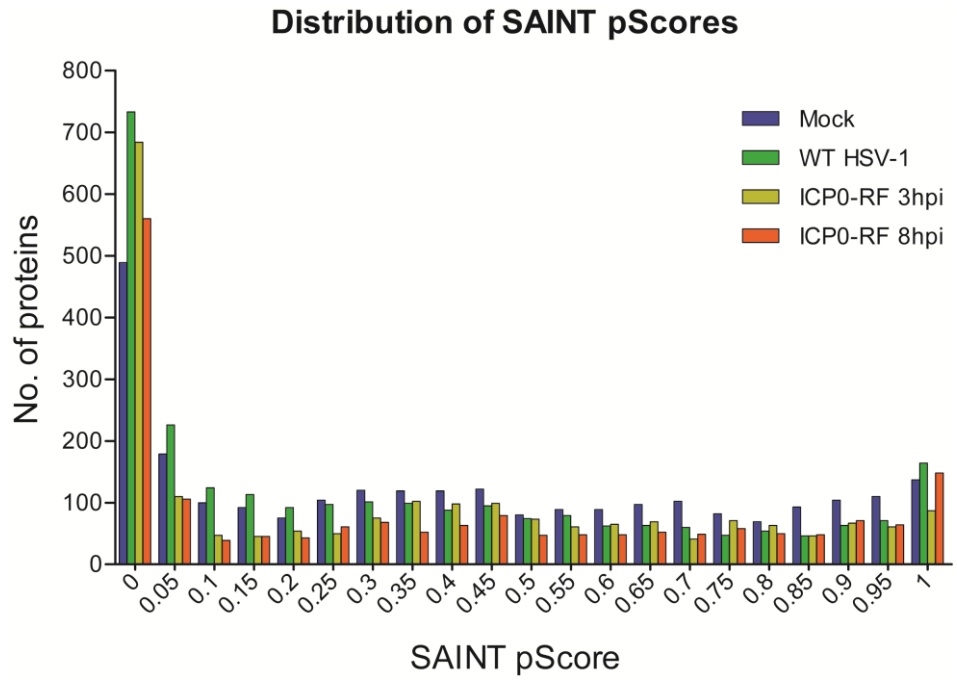
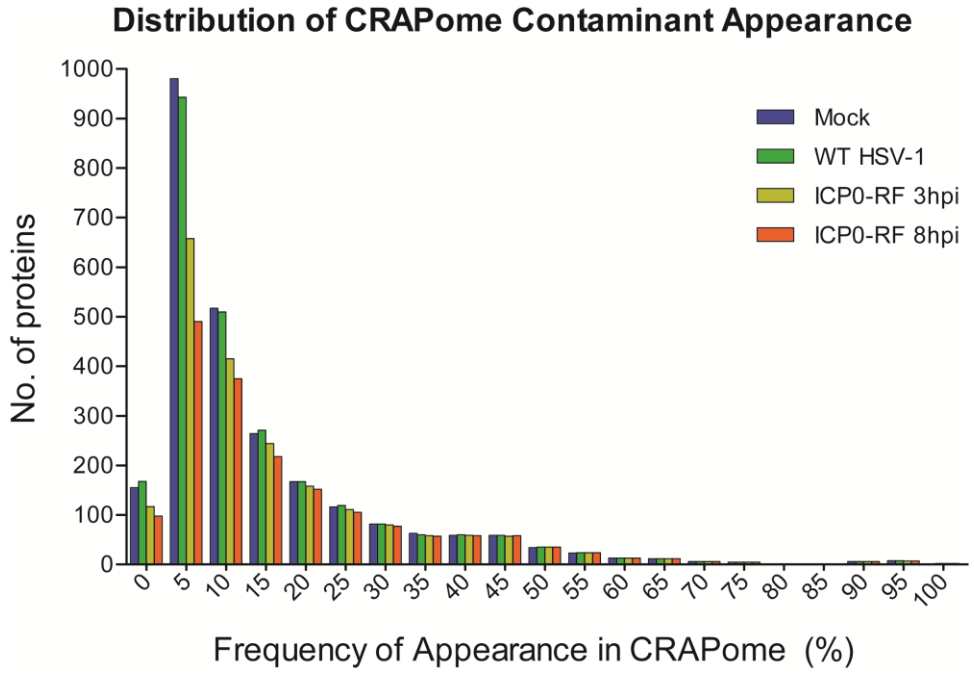

STK3 SNX6 ZNF574 UBTF ZNF512 PWP1 RPL36A UTP3 APHOSPH ESF1
UPF1 CENPV HP1BP3 DHX33 H2AFY2 CEBPZ ASCC3 PHF14 PCID2 ZNF638
LZTR1 CENPB MBD2 CIRH1A H2AFY DYNLL1 KAT7 KHDRBS3 USP36 RBM14
CARM1 ZNF579 WHSC1 REXO4 BRD1 ADD1 POLDIP3 CNOT1 RBM15 SMCHD1
UL25 UL26 UL36 UL17 UL19 UL38 UL6 UL37 UL21
TK UL5 UL42 UL30 US1 UL47 UL46 UL13 ICP0 US2
IFI16
CBX5 SP100 MKI67 KIF23 CTNND1 CBX1 CBX3 SUMO1 TOP2A RACGAP1 CDC42 TOP2B
RRP8 BAZ1B SMARCC2 BAZ2A SMARCA5 SMARCA1 ATAD2 CDK1 INCENP
WDR18 PELP1 SENP3 TEX10 LAS1L
RFC3 RFC2
NUP85 NUP214 NXF1 RBM8A NUP54 SEH1L NUP205 RAE1 NEMF NUP93 POM121C NUP50 RGPD8 NUP98 NDC1 NUP160 NUP62 NUP153 MCM3AP NCBP1 GLE1 NUP43 NUP133 SEC13 NUP37 NUP88 NUPL1 NUP35 AAAS SRRM1 NUP188 AHCTF1 NUP107
SUN2 SYNE1 SUN1
LEMD2 TOR1AIP1 NUMA1
WDR61 PAF1 SUPT5H SSRP1 SUPT16H XAB2 POLR2A TCEB1 CTCF
LRRFIP2 LRRFIP1 FLII
CSNK2A2 CSNK2B
TRIM56 TRIM26 USP7
DHX36 ZC3HAV1 FBXW11
APOBEC3C DHX30 UACA
APOBEC3B NKRF PRKRA
TRIP12 ACIN1 PNN SAP18
PIP4K2A GLYR1 NMNAT1
DHX29 LARP4B SRP72 FXR2 FXR1 FMR1 SRP68 KIAA0020 EIF6 LARP1 AGO2
UTP18 TBL3 PWP2 AATF DDX47 NOC2L PHIP WDR12 WDR3 UTP6 BOP1 PES1 WDR36
PAK1IP1 DDX24 GTPBP4 RCL1 RRP1B NOP16 KRI1 BMS1 RRS1 UTP15 NLE1 UTP14A FCF1 UTP11L WDR46 HEATR1 URB1
LARP7 RRP9 GNL3 DDX18 CCDC86 MRTO4 RRP12 NOL10 PDCD11 MPHOSPH6 NOL6 IMP4 NOC4L DDX56 UTP20 PPAN
TSR1 DDX27 MAK16 LYAR KRR1 NOL11 DDX54 MKI67IP NAT10 RPP30 BYSL BRIX1 DDX52 DCAF13 DDX10 EMG1
POP1 RPF2 BNA1BP2 BM34 RBM19 NOC3L NSA2 NOL9 URB2 GNL2 RSL24D1 RRP1 WDR75 NVL WDR74 RPF1
CRNKL1 NHP2 NHP2L1 COIL DDX28 RBM28
PRPF38A SRPK1 DHX8 RBM25 PRPF40A SART1
SART3 PLRG1 SNRNP27 YTHDC1 WTAP SON
DHX16 POP4
EDC4 XRN1 EXOSC3 ZC3H14 EXOSC10 EXOSC4 EXOSC7 EXOSC2 EXOSC8
Interactions
Mock
HSV-1
Both
Spectral Enrichment Log2(HSV-1/Uninf.)
Uninf.
HSV-1

Figure S2.


IFI16
UL47 UL46 UL14
US1 UL48
UL13 ICP4
ICP0
UL36
UL21 UL25
UL17 UL37
UL38
UL31 US3
UL51 UL7
US10 UL56
TK UL40
UL50 UL39
ICP8 UL12 UL42
US11 US2
UL54 UL41
UL24
gD gM
gL gH
gB
DDX60L ACADVL
DDX28 MKI67
GLYR1 PRKAR1A
NMNAT1
CUL4A DDX47 CSNK2A2 LGALS7 ADD1
PPP2CB PHIP H1F0 PDCD6IP FGF2
FHL2 FXR1 GSDMA
DNAJC10 TJP1
IFIT2 ADAR LRRFIP2 IPO7
SQSTM1 PRKRA FLII EIF2AK2
TRIM26 CCDC86 APOBEC3C
LRRFIP1 DYNLL1
AATF
NOC2L
SUMO1
PML
HLA-A HLA-C
KIF23
USP7
CBX1 CBX3
SP100
ATAD2
SSRP1 ZNF512
BAZ1B
RNF213
SMARCA1
HUWE1 DCAF13
SMARCC2 BAZ2A
BTF3 PSPC1 POLR2B HELZ2
PDAP1
CSNK2B NLE1 WHSC1 OSBPL8
SMARCA5 TOP2A HP1BP3
PELP1 SNX6 PAF1 POLR2A ASCC3
RBM19
LEMD2 RAB14
PBRM1 SMCHD1 ACIN1
TEX10 LAS1L NKRF CEBPZ PWP1 LZTR1
NUMA1
SDCBP CDC42
SRP72
SUN2
GTPBP4 PPHLN1 PTPLAD1 MAPK3
KIAA0020 FXR2
POLDIP3
SUN1 SYNE1
TNKS1BP1 WTAP
STAU1
PRPF40A
RSL24D1 PWP2 DDX52 DKC1 UTP6 BYSL
TMEM43 CEP170
SON
PLRG1
NOC4L RRP12 TBL3 NHP2 NOL6 GNL3 DDX51
NUP153
SAP18
WDR75 MRTO4 UTP18 RPP30 NVL WDR43 RCL1
NUP88 NUP37 NUP188 NDC1
SNRNP27
NOL10 BRIX1 RPF2 RRP9 LARP7 KRI1 DDX24
AHCTF1 NUP214 NUP93 CKAP5
PRRC2A
DHX30
FTSJ3 NSA2 RRP1B DDX56 NGDN GRWD1 MPHOSPH10
NUP133 RAE1 NUP85 GLE1
POP1
EXOSC6
WDR36 RBM34 DDX27 UTP15 WDR46 WDR12 SKIV2L2
NUP98 NUP62 AAAS RGPD8
POM121C
EXOSC10
WDR3 EIF3K BOP1 MKI67IP URB1 NOP16 PDCD11
SEH1L NUP205 MCM3AP NXF1
EDC4
RBM28
EXOSC4
IMP4 NAT10 CIRH1A HEATR1 DDX18 NOL11 DNTTIP2
Spectral Enrichment
Log2(8 hpi / 3 hpi)
3 hpi
8hpi
Interactions
ICP0-RF
Both ICP0-RF and Mock and/or Uninf.


Distribution of SAINT pScores
No. of proteins
800
700
600
500
400
300
200
100
0
Mock
WT HSV-1
ICP0-RF 3hpi
ICP0-RF 8hpi
0
0.05
0.1
0.15
0.2
0.25
0.3
0.35
0.4
0.45
0.5
0.55
0.6
0.65
0.7
0.75
0.8
0.85
0.9
0.95
1
SAINT pScore


Distribution of CRAPome Contaminant Appearance
No. of proteins
1000
900
800
700
600
500
400
300
200
100
0
Mock
WT HSV-1
ICP0-RF 3hpi
ICP0-RF 8hpi
0
5
10
15
20
25
30
35
40
45
50
55
60
65
70
75
80
85
90
95
100
Frequency of Appearance in CRAPome (%)

Supplementary Figure Legends

Figure S1. Complete interaction network of endogenous IFI16 in uninfected HFF cells and after infection with *WT* HSV-1. SAINT- and CRAPome-filtered interactions with IFI16 were assembled using STRING and visualized as a network in Cytoscape. Node shape indicates the presence or absence of a protein in other IP conditions. Node colors indicate Log_2-transformed normalized spectral count fold changes between uninfected and *WT* HSV-1 infected cells.

Figure S2. Complete interaction network of endogenous IFI16 after infection with ICP0-RF at 3 and 8 hpi. SAINT- and CRAPome-filtered interactions with IFI16 were assembled using STRING and visualized as a network in Cytoscape. Node shape indicates the presence or absence of a protein in other IP conditions. Node colors indicate Log_2-transformed normalized spectral count fold changes between 3 and 8 hpi conditions.

Figure S3. Distribution analysis of average SAINT probability scores and CRAPome contaminant frequency for all interactions detected within each IFI16 immunoaffinity purification condition. The distributions of the averages of the top two SAINT scores across three biological replicates for all identified endogenous IFI16 interactions detected in primary fibroblasts for each tested condition are shown (top). Using the 411 total control immunoaffinity isolation experiments submitted to the CRAPome database, percent frequencies of appearance for all identified endogenous IFI16 interactions identified by mass spectrometry within each tested condition were calculated. Their distribution is displayed (bottom).

Supplementary Table Legends

Table S1. IFI16 interactions that passed specificity criteria in uninfected HFFs, or after infection with WT HSV-1. (A) Complete list of IFI16 interactions that passed all the specificity criteria in uninfected HFFs and cells infected with WT HSV-1. (B-C) List of IFI16 interactions that passed SAINT filtering, but not yet the additional filtering based on CRAPome and known cytoskeletal, extracellular, and other abundant sticky proteins; B – uninfected cells; C - cells infected with WT HSV-1. (D) Protein amino acid sequence coverages, illustrated as percentages. (E) Number of unique peptides for all identified proteins.

Table S2. IFI16 interactions that passed specificity criteria in HFFs after infection with *ICP0-RF* at 3 or 8 hpi. (A) Complete list of IFI16 interactions that passed all the specificity criteria in cells infected with ICP0-RF HSV-1. (B-C) List of IFI16 interactions that passed SAINT filtering, but not yet the additional filtering based on CRAPome and known cytoskeletal, extracellular, and other abundant sticky proteins; B – 3 hpi; C – 8 hpi. (D) Protein amino acid sequence coverages, illustrated as percentages. (E) Number of unique peptides for all identified proteins.

Table S3. List of associations excluded as likely non-specific based on CRAPome. Following SAINT filtering, IFI16 interactions were further compared to negative control samples from the web-based CRAPome repository. Proteins with > 20% appearance within the 411 deposited control experiments, as well as common non-specific proteins were filtered out prior to final network analysis.

Table S4. Oligonucleotides for qPCR and shRNA. DNA primer sequences for RT-qPCR of cytokines and oligonucleotide sequences for shRNA constructs.